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A New Species of *Microhyla* (Amphibia: Anura: Microhylidae) from Langbian Plateau, Central Vietnam

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Abstract We describe a new species, *Microhyla hongiaoensis* **sp. nov.**, from Lam Dong Province, southern Vietnam based on morphological data and molecular evidences. The new species is sister to *M. pulchella* by molecular phylogenetics and also most closely resembles *M. pulchella* in morphological characteristics, albeit differs from its congeners by a combination of the following morphological features: (1) size medium (SVL 13.6–14.7 mm in males and 18.3–18.6 mm in females); (2) fingers II–IV with small disks, dorsal surface of disks, without median longitudinal groove; (3) webbing formula I1½ – 2II1 – 2III1 – 2½IV2¼ – 1V; (4) toe disks with dorsal median longitudinal groove; (5) dorsal back without two small black spots; (6) one small black spot adjacent behind the eyes; (7) few small black scapular spots in the flanks-belly and inguinal region; (8) palm with two small metatarsal tubercles; (9) tibiotarsal reaching beyond snout. *M. hongiaoensis* **sp. nov.** occurs in evergreen montane tropical forests at an elevation of 1500 m a.s.l.

Keywords new species, *Microhyla*, Bi Doup-Nui Ba, national park, morphology, molecular phylogeny

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1. Introduction

The genus *Microhyla* Tschudi, 1838 is an assemblage of generally small, mostly ground-dwelling frogs. The genus currently contains 50 species with a distribution range from the Ryukyu Archipelago (Japan) and eastern China westwards to the northern part of India, Sri Lanka and mainland Southeast Asia, and southwards to Indonesia (Frost, 2019). Remarkably, 23 new species of *Microhyla* have been described during the last decade (Frost, 2019) and recent molecular studies have discovered numerous highly divergent lineages and cryptic species (Matsui *et al*., 2013; Hasan *et al*., 2015; Howlader *et al*., 2015; Seshadri *et al*., 2016; Wijayathilaka *et al*., 2016; Yuan *et al*., 2016; Khatiwada *et al*., 2017; Zhang *et al*., 2018; Garg *et al*., 2019; Li *et al*., 2019; Nguyen *et al*., 2019; Poyarkov *et al*., 2014, 2019).

In Vietnam, 17 species of *Microhyla* have been recorded so far (Frost, 2019) and the greatest diversity of the genus occurs in the central and southern parts of the Truong Son Range (also known as the Central Highlands or the Tay Nguyen Plateau) (Figure 1). Tay Nguyen Plateau, including Langbian Plateau, harbors the highest diversity of amphibians in Indochina with a high level of local endemism and new species discovery (Bain and Hurley, 2011; Geissler *et al*., 2015; Chen *et al*., 2018; Nguyen *et al*., 2019). This region appears to be one of the centers of radiation for the genus (Poyarkov *et al*., 2014). The following 14 species of *Microhyla* have been recorded

from the Tay Nguyen Plateau: *M. annamensis* Smith, 1923; *M. aurantiventris* Nguyen, Poyarkov, Nguyen, Nguyen, Tran, Gorin, Murphy, and Nguyen, 2019; *M. berdmorei* (Blyth, 1856) Bain and Nguyen, 2004; *M. butleri* Boulenger, 1900; *M. darevskii* Poyarkov, Vassilieva, Orlov, Galoyan, Tran, Le, Kretova, and Geissler, 2014; *M. fusca* Andersson, 1942; *M. heymonsi* Vogt, 1913; *M. minuta* Poyarkov, Vassilieva, Orlov, Galoyan, Tran, Le, Kretova, and Geissler, 2014; *M. mukhlesuri* Hasan, Islam, Kuramoto, Kurabayashi, and Sumida, 2014; *M. nanapollexa* Bain and Nguyen, 2004; *M. pineticola* Poyarkov, Vassilieva, Orlov, Galoyan, Tran, Le, Kretova, and Geissler, 2014; *M. pulchella* Poyarkov, Vassilieva, Orlov, Galoyan, Tran, Le, Kretova, and Geissler, 2014; *M. pulchra* (Hallowell, 1861) Bain and Nguyen, 2004; *M. pulverata* Bain and Nguyen, 2004 (Inger *et al*., 1999; Bain and Nguyen 2004; Hoang *et al*., 2013; Poyarkov *et al*., 2014; Nguyen *et al*., 2019).

During our recent field surveys in Langbian Plateau of Lam Dong Province in 2018, we collected a series of adult microhylid frogs that morphologically resemble *M. pulchella,* a species that was recently described from Bi Doup-Nui Ba National Park in Lam Dong Province (Poyarkov *et al*., 2014). However, these specimens are smaller in size than any known specimen of *M. pulchella* and have greyish-brown to light-brown dorsum with dark-brown markings extending from their interorbital bar to hindlimb, forming a double-waisted figure usually in a light-brown color. Furthermore, they were reproducing syntonically with *M. pulchella* that differed markedly from those of sympatric *M. pulchella*. Subsequent analyses based on morphological and molecular data showed that these specimens represented an independent evolutionary lineage that could not be assigned to any known species of *Microhyla*. We herein describe the population of *Microhyla* from Lam Dong province as a new species.

2. Materials and Methods

2.1. Sampling Field surveys were conducted in Bi Doup-Nui Ba National Park, Lam Dong Province, Vietnam (Figure 1) in May 2018 by C. V. Hoang, A. M. Luong, Y. T. Nguyen, N. L. Orlov, L. Iogansen (hereafter C. V. Hoang *et al*.). Geographic coordinates and elevation were obtained using a Garmin GPSMAP 78S (WGS 84 data). After photographing the specimens alive, they were euthanized in a closed vessel with a piece of cotton wool containing ethyl acetate (Simmons, 2002), fixed in 80% ethanol for five hours, and then transferred to 70% ethanol for permanent storage. Tissue samples were preserved separately in 70% ethanol prior to fixation. Eleven specimens of the new form (Tables 1, 3) were collected and subsequently deposited in the collection of the Vietnam National Museum of Nature (VNMN 07385, 07388, 07390, 07477), Institute of

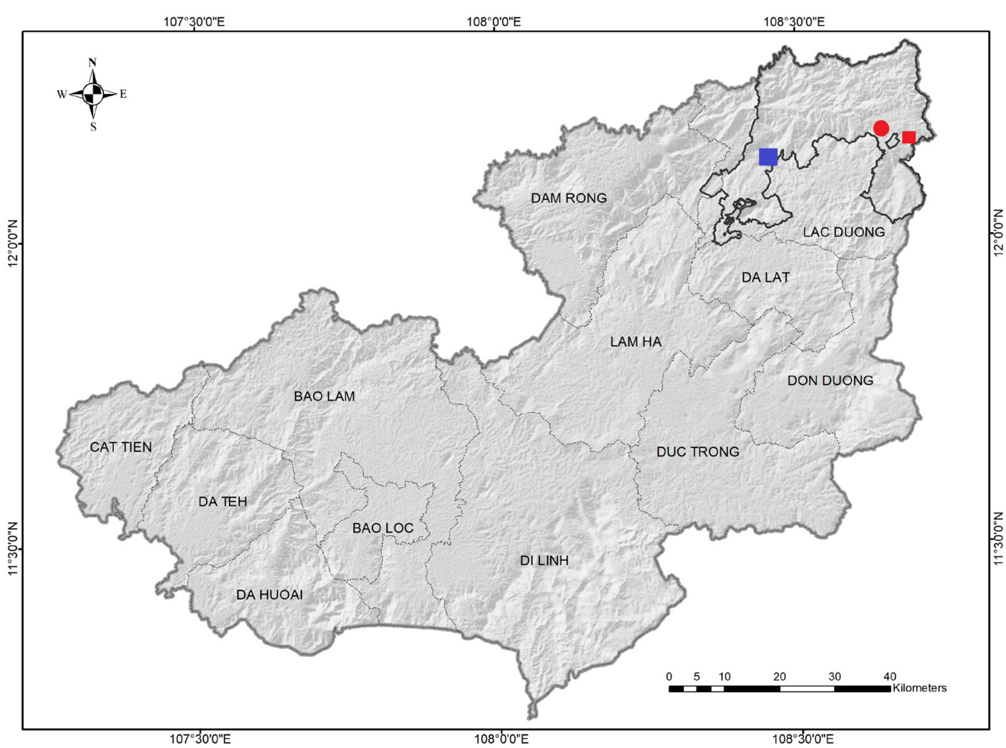

Figure 1 Map showing the type locality (red circle) of *Microhyla hongiaoensis* **sp. nov.** in Lam Dong Province, Vietnam (12°11'18.3"N, 108°40'29.0"E); blue square is a known distribution point of *M. pulchella* (12°5'55.81"N, 108°21'26.83"E); red square is a known distribution point of *M. hongiaoensis* **sp. nov.** and *M. pulchella* (12°10'27.44"N, 108°42'24.64"E).

Table 1 Specimens and sequences of *Microhyla* and outgroup Microhylidae representatives used in molecular analyses.

No.	Species	Locality	Voucher No.	GenBank No.	Reference
1	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	CIB-VNMN 07617	MN475176	This study
2	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	CIB-VNMN 07483	MN475177	This study
3	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07477	MN475178	This study
4	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	IEBR4576	MN475179	This study
5	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	IEBR4574	MN475180	This study
6	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	IEBR4573	MN475181	This study
7	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	IEBR4575	MN475182	This study
8	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	CIB-VNMN 07448	MN475183	This study
9	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07390	MN475184	This study
10	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07388	MN475185	This study
11	*M. hongiaoensis* **sp. nov.**	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07385	MN475196	This study
12	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07343	MN475186	This study
13	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07618	MN475187	This study
14	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07614	MN475188	This study
15	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07581	MN475189	This study
16	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07531	MN475191	This study
17	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07574	MN475190	This study
18	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07478	MN475192	This study
19	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07391	MN475193	This study
20	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07389	MN475194	This study
21	*M. pulchella*	Viet Nam: Lam Dong, Bi Doup Nui Ba	VNMN 07387	MN475195	This study
22	*M. achatina*	Indonesia:Java, Ungaran	MZB Amp 16402	AB634656	Matsui *et al.*, 2011
23	*M. aurantiventris*	Vietnam: Gia Lai, Tram Lap	ITBCZ4361	MH286426	Nguyen *et al.*, 2019
24	*M. annectens*	Malaysia: Selangor, Genting	KUHE53373	AB634658	Matsui *et al.*, 2011
25	*M. beilunensis*	China: Zhejiang, Ningbo, Beilun	CIB BL002	MH234535	Zhang *et al.*, 2018
26	*M. berdmorei*	Myanmar: Sagaing, Alaungdaw Kathapa National Park	CAS:HERP:204876	KC179981	de Sa *et al.*, 2012; Garg *et al.*, 2019
27	*M. borneensis*	Malaysia: Sarawak, Gunung Serapi	KUHE53165	AB598329	Matsui, 2011
28	*M. butleri*	Vietnam: A Roang, A Luoi	KUHE40591	AB634664	Matsui *et al.*, 2011
29	*M. chakrapanii*	India: Andaman Islands	Not preserved	MH807389	Garg *et al.*, 2019
30	*M. darreli*	India: Thiruvananthapuram, Karamana	ZSI/WGRC/V/A/962	MH807390	Garg *et al.*, 2019
31	*M. fissipes*	China: Anhui, Huangshan	KUHE32943	AB201185	Matsui *et al.*, 2005
32	*M. gadjahmadai*	Indonesia, Sumatra	MZB Amph 15292	MK034333	Atmaja *et al.*, 2018
33	*M. heymonsi*	China: Sichuan, Zihuai	NA	AF215372	Vences, 2000
34	*M. karunaratnei*	Sri Lanka: Morningside, Sinharaja	DZ 1529	MH807392	Garg *et al.*, 2019
35	*M. kodial*	India: Karnataka, Mangalore	NCBS-AY588	MF919453	Vineeth *et al.*, 2018
36	*M. laterite*	India: Karnataka, Manipal	BNHS 5965	KT600670	Seshadri *et al.*, 2016; Garg *et al.*, 2019
37	*M. malang*	Malaysia: Sarawak, Gunung Serapi	KUHE32943	AB598319	Matsui, 2011
38	*M. mantheyi*	Malaysia: Selangor, Templer Park	KUHE15726	AB598333	Matsui, 2011
39	*M. marmorata*	Vietnam: Kon Tum	KP-MD2018.24	MN453610	This study
40	*M. mihintalei*	Sri Lanka: Anuradhapura	DZ1468	KU214861	Garg *et al.*, 2019
41	*M. mixtura*	China: Shaanxi	CIBZMH2017061201	MH234532	Zhang *et al.*, 2018
42	*M. mukhlesuri*	Bangladesh: Chittagong, Raozan	IABHU3879	AB543608	Garg *et al.*, 2019
43	*M. mymensinghensis*	Bangladesh: Mymensingh	IABHU F5012 1-6	AB530529	Garg *et al.*, 2019
44	*M. nanapollexa*	Vietnam: Quang Nam	PT-484	KM509164	Garg *et al.*, 2019
45	*M. nilphamariensis*	Bangladesh: Nilphamari	MZH-2362	KP072789	Garg *et al.*, 2019
46	*M. okinavensis*	Japan: Okinawa	IABHU5263	AB303950	Garg *et al.*, 2019
47	*M. orientalis*	Indonesia: Bali, Wongaya Gede	KUHE55073	AB781469	Garg *et al.*, 2019
48	*M. ornata*	India: Andhra Pradesh, Maredumilli	SDBDU 2015.2898	MH549619	Garg *et al.*, 2019

(Continued Table 1)

No.	Species	Locality	Voucher No.	GenBank No.	Reference
49	*M. palmipes*	Indonesia: Bali	MZB Amp 16255	AB634670	Garg *et al.*, 2019
50	*M. perparva*	Malaysia: Sarawak	KUHE53675	AB634673	Garg *et al.*, 2019
51	*M. petrigena*	Malaysia: Sarawak	KUHE53743	AB634675	Garg *et al.*, 2019
52	*M. pulchra*	China: Guangdong	NA	KF798195	Garg *et al.*, 2019
53	*M. rubra*	India: Karnataka	NA	AB201192	Garg *et al.*, 2019
54	*M. sholigari*	India: Talapu, Madikeri	RBRL 060709-29	AB530631	Garg *et al.*, 2019
55	*M. superciliaris*	Malaysia: Negeri Sembilan, Kenaboi	KUHE53371	AB634683	Garg *et al.*, 2019
56	*M. taraiensis*	Nepal: Mechi, Jhapa, Jamun Khadi	JRK201525	KY655952	Garg *et al.*, 2019
57	*M. zeylanica*	Sri Lanka: Horton plains	DZ 1419	MH807428	Garg *et al.*, 2019
	Outgroup				
58	*Kaloula pulchra*	Thailand: Kanchanaburi	KUHE35171	AB201194	Garg *et al.*, 2019

Ecology and Biological Resources (IEBR 4573–4576), Hanoi, Vietnam. For comparative specimens, 21 specimens of *Microhyla pulchella* (Table 3) were collected in Bi Doup-Nui Ba National Park, Lam Dong Province, Vietnam (VNMN 07478, 07718, 07710, 07389, 07713–07714, 07391, 07693, 07705–07708, 07683, 07581, 07694, 07386, 07437, 07531, 07691, 07614, 07574). Three specimens of the new form (VNMN 07385, 07388, 07390) and two specimens of *Microhyla pulchella* (VNMN 07389, 07391) were collected in the same breeding pond. Sex was determined by direct observation of calling males or by gonadal dissection after euthanasia.

2.2. Molecular analyses

DNA extraction and sequencing In this study, we amplified a 1936 base pair (bp) length fragment of the 12S rRNA–16S rRNA mitochondrial gene and the complete sequence of tRNAVal that was used recently for *Microhyla* (Nguyen *et al.*, 2019). DNA of 21 tissue samples were extracted using TIANamp Genomic DNA kit (TIANGEN BIOTECH, Beijing, China), *Tiangen following the manufacturers' instructions* (Table 1)*. Total DNA was* amplified using an Eppendorf PCR machine. PCR total volume was 25 µL, consisting of 12 µL of Mastermix, 6 µL of water, 1 µL of each primer at a concentration of 10 pmol/µL, and 5 µL of DNA. Primers used in the PCR and sequencing were as follows: 12SAL (5'-AAACTGGGATTAGATACCCCA CTAT-3'; forward), 16S2000H (5'-GTGATTAYGCTACCTT TGCACGGT-3'; reverse) (Zhang *et al., 2008)* and LR-N-13398 (5'-CGCCTGTTTACCAAAAACAT-3'; forward), LR-J 12887 (5'-CCGGTCTGAACTCAGATCACGT -3'; reverse) (Simon, 1994). PCR conditions: 94°C for 5 minutes of initial denaturation; with 35 cycles of denaturation at 94°C for 30 s, annealing at 56°C for 30 s, and extension at 72°C for 45 s; and the final extension at 72°C for 7 minutes. PCR products were sent to Tsingke Biological Technology company for sequencing (http://www.tsingke.net). The obtained sequences were deposited in GenBank under the accession numbers MN475176–475196 (Table 1).

Phylogenetic analysis In addition to 11 sequences of the new form from Bi Doup-Nui Ba National Park and 10 sequences of *M. pulchella* from newly collected samples, we used 35 available sequences of 12S rRNA–16S rRNA in GenBank (Garg *et al.*, 2019) and a sequence (MN453610) of *M. marmorata* from Kon Tum Province, Vietnam, for phylogenetic analyses of the genus *Microhyla*. Sequences of *Kaloula pulchra* were included in the analysis as the outgroup (Van Bocxlaer *et al.*, 2006). Locality information and accession numbers for all sequences included in the analysis can be found in Table 1.

Phylogenetic trees were constructed by using maximum likelihood (ML) and Bayesian inference (BI) analyses. Chromas Pro software (Technelysium Pty Ltd., Tewantin, Australia) was used to edit the sequences, and then aligned using the ClustalW (Thompson *et al.*, 1997) option in MEGA 7.0 (Kumar *et al.*, 2016) with default parameters and subsequently optimized manually in BioEdit 7.0.5.2 (Hall, 1999). We then checked the initial alignments by eye and adjusted slightly. Prior to ML and Bayesian tests, phylogenetic analyses were performed in MrBayes 3.2 (Ronquist *et al.*, 2012). We chose the optimum substitution models for entire sequences using Kakusan 4 (Tanabe, 2011) based on the Akaike information criterion (AIC). The best model selected for ML was the general time reversible model (GTR: Tavaré 1986) with a gamma shape parameter (G: 0.232 in ML and 0.249 in BI). The BI summarized two independent runs of four Markov Chains for 10,000,000 generations. A tree was sampled every 100 generations and a consensus topology was calculated for 70,000 trees after discarding the first 30,001 trees (burn-in = 30 001) (Nguyen *et al.*, 2017). We checked parameter estimations and convergence using Tracer version 1.5 (Rambaut and Drummond, 2009). The strength of nodal support in the ML tree was analyzed using non-parametric bootstrapping (MLBS) with 1000 replicates. We regarded tree nodes in the ML tree with bootstrap values of 75% or greater as sufficiently resolved (Hillis and Bull, 1993; Huelsenbeck and Hillis, 1993), and nodes with a BPP of 95% or

greater as significant in the BI analysis (Leaché and Reeder, 2002).

2.3. Morphological analysis

Specimens examined A total of 32 specimens of *Microhyla* was collected from Bi Doup-Nui Ba National Park, Lac Duong District, Lam Dong Province, in May 2018 (Table 3).

Measurements Measurements were taken from preserved specimens by V.C. Hoang with a digital caliper to the nearest 0.1 mm under a stereo microscope (Table 3). The following morphological characteristing were used (see Matsui, 2011; Matsui *et al*., 2013; Poyarkov *et al*., 2014): (1) snout-vent length (SVL, from the tip of snout to cloaca); (2) head length (HL, from tip of snout to posterior margin of jaw angle); (3) snout length (SL, from the anterior corner of eye to the tip of snout); (4) eye length (EL, the distance between the anterior and posterior corners of the eye); (5) nostril-eye length (NEL, the distance between the anterior corner of the eye and the nostril); (6) head width (HW, the maximum width of the head on the level of mouth angles in ventral view); (7) internarial distance (IND); (8) interorbital distance (IOD, the shortest distance between the medial edges of eyeballs in dorsal view); (9) upper eyelid width (UEW, the widest distance from the medial edge of eyeball to the lateral edge of the upper eyelid); (10) forelimb length (FLL, length of straightened forelimb to tip of third finger); (11) lower arm and hand length (LAL, distance from elbow to tip of third finger); (12) hand length (HAL, from proximal end of outer palmar [metacarpal] tubercle to tip of third finger); (13) inner palmar tubercle length (IPTL, maximal distance from proximal to distal ends of inner palmar tubercle); (14) outer palmar tubercle length (OPTL, maximal diameter of outer palmar tubercle); (15) hindlimb length (HLL, length of straightened hindlimb from groin to tip of fourth toe); (16) tibia length (TL, the distance between the knee and tibiotarsal articulation); (17) foot length (FL, from distal end of tibia to tip of toe IV); (18) inner metatarsal tubercle length (IMTL, maximal length of inner metatarsal tubercle); (19) first toe length (1TOEL, from distal end of inner metatarsal tubercle to tip of first toe); (20) outer metatarsal tubercle length (OMTL); (21) first finger width (1FW, measured at the distal phalanx); (22–25) finger lengths (1–3FLO, 2–4FLI; for outer side (O) of the first, inner side (I) of the fourth, and both sides of the remaining fingers, measured between tip and the junction of the neighbouring finger); (26–28) finger disk diameters (2–4FDW); and (29–33) toe disk diameters (1–5TDW). Terminology for describing eye coloration in life followed Glaw and Vences (1997); webbing formula followed Savage (1975).

Morphological comparisons Morphological comparisons were based on 50 specimens examined and data from literatures (Tables 3, 4): Boulenger (1897, 1900); Smith (1923); Parker (1928, 1934); Andersson (1942); Bourret (1942); Parker and Osman (1948); Pillai (1977); Inger and Frogner (1979); Inger (1989); Dutta and Ray (2000); Bain and Nguyen (2004); Das *et al*. (2007); Das and Haas (2010); Fei *et al*. (2012); Matsui (2011); Matsui *et al*. (2013); Hasan *et al*. (2014); Poyarkov *et al*. (2014); Howlader *et al*. (2015); Seshadri *et al*. (2016); Wijayathilaka *et al*. (2016); Khatiwada *et al*. (2017); Zhang *et al*. (2018); Nguyen *et al*. (2019); Garg *et al*. (2019); Poyarkov *et al*. (2019); and Li *et al*. (2019).

Principal component analysis (PCA) Measurement data were size-corrected and then were used to compare the morphometric difference between eight males and three females of the new form vs. the male holotype, 17 males and four females of *M. pulchella* from Bi Doup-Nui Ba National Park. All statistical analyses were performed using PAST 2.17b software (Hammer *et al*., 2001).

3. Results

3.1. Molecular systematics

Sequence variation In the final alignment of 12S rRNA–16S rRNA, 993 sites were conserved and 964 sites exhibited variation, of which 557 were found to be potentially parsimony-informative. The transition-transversion bias (R) was estimated as 1. Nucleotide frequencies were A = 32.20%, T = 23.15%, C = 23.85%, and G = 20.81% (data for ingroup only).

Interspecific uncorrected p-distance In the *Microhyla* species group, the uncorrected p-distance ranged from 1.7% (between *M. mixtura* and *M. okinavensis*) to 16.4% (between *M. annectens* and *M. zeylanica*) (Table 2). The genetic divergence of the new form and its congeners ranged from 2.8% (compared with *M. pulchella*) to 13.2 % (compared with *M. zeylanica*). These values were higher than those between some other recognized species of *Microhyla* (2.6% between *M. karunaratnei* and *M. darreli*, *M. borneensis* and *M. malang*, *M. mukhlesuri* and *M. fissipes*; 1.9% between *M. okinavensis* and *M. beilunensis*; 1.7% between *M. okinavensis* and *M. mixtura*). The new form is separated from *M. pulchella*, the most closely related species in morphology, by a genetic distance of approximately 2.8% based on 12S–16S rRNA mtDNA fragments (Table 2). It is noted that three specimens of the new form (VNMN 07385, 07388, 07390) and two specimens of *Microhyla pulchella* (VNMN 07389, 07391), collected from the same breeding pond, are also distinguished from each other by a genetic difference of at least 2.8%.

Phylogenetic relationships The BI and ML analyses produced topologies with −lnL = 13118.5911 and 13107.9365, respectively. BI and ML analyses obtained similar topologies (Figure 2) that differed only at several poorly supported basal nodes. Our matrilineal genealogy was consistent with those of Matsui *et al*. (2011), Peloso *et al*. (2016) and Garg *et al*. (2019).

The BI genealogy inferred the following set of phylogenetic relationships: Monophyly of Microhylinae was not supported and the relationships among microhylid subfamilies remained unresolved (Nguyen *et al*., 2019) (Figure 2). In both analyses, the newly collected *Microhyla* form was recovered as sister group to *M. pulchella* (Poyarkov *et al*., 2014) from Lam Dong Province, Langbian plateau, Vietnam with high nodal support values (1.0/99.7).

3.2. Morphological analysis The new form of *Microhyla* from Bi Doup-Nui Ba National Park, Lam Dong Province, Vietnam, is compared with all known congeners as following.

The new form of *Microhyla* from Lam Dong Province differs from other species of *Microhyla* by having a smaller size, slender habitus; bluntly round snout; skin on dorsum scattered by small tubercles; finger I shorter than one-half the length of finger II; tips of three outer fingers (II–IV) weakly dilated, forming weak disks, largest on finger III, without dorsal median longitudinal grooves; tips of all toes distinctly dilated into wide disks with narrow peripheral grooves; and tibiotarsal articulation reaching

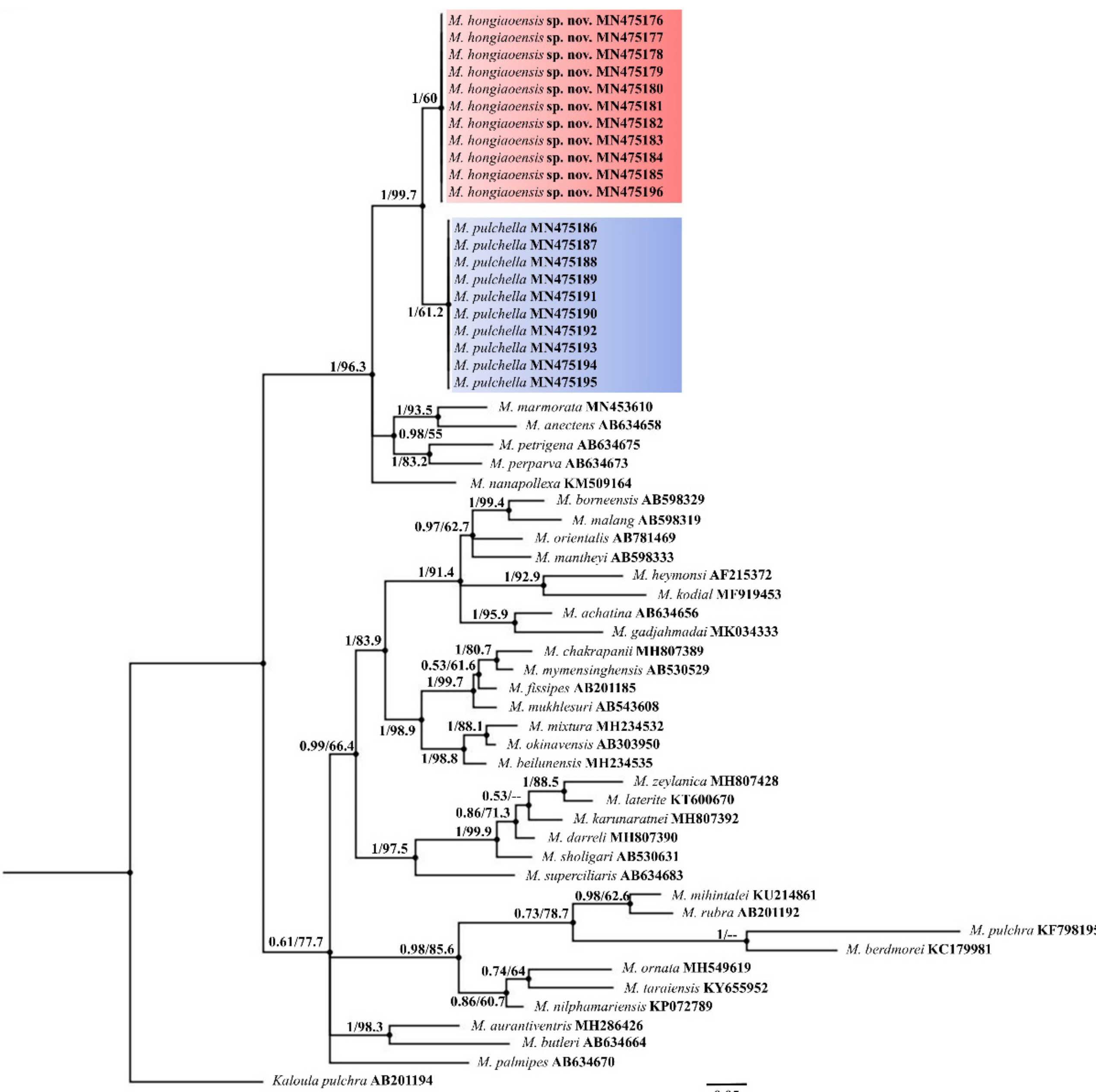

Figure 2 Bayesian inference matrilineal genealogy of *Microhyla* derived from the analysis of 12S rRNA–16S rRNA mtDNA sequences. Numbers above and under branches are Bayesian posterior probabilities and ML bootstrap values.

Table 2 Uncorrected (“*p*”) distance matrix showing percentage pair wise genetic divergence 12S rRNA–16S rRNA between members of the *Microhyla* species group.

	Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	*M. hongiaoensis* **sp. nov.**																				
2	*M. hongiaoensis* **sp. nov.**	0.000																			
3	*M. hongiaoensis* **sp. nov.**	0.000	0.000																		
4	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000																	
5	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000																
6	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000	0.000															
7	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000	0.000	0.000														
8	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000	0.000	0.000	0.000													
9	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000												
10	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000											
11	*M. hongiaoensis* **sp. nov.**	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000										
12	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028									
13	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000								
14	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000							
15	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000						
16	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000	0.000					
17	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000	0.000	0.000				
18	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000	0.000	0.000	0.000			
19	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000	0.000	0.000	0.000	0.000		
20	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	
21	*M. pulchella*	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.028	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
22	*M. marmorata*	0.065	0.065	0.065	0.065	0.065	0.065	0.065	0.065	0.065	0.065	0.065	0.082	0.082	0.082	0.082	0.082	0.082	0.082	0.082	0.082
23	*M. zeylanica*	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.135	0.135	0.135	0.135	0.135	0.135	0.135	0.135	0.135
24	*M. karunaratnei*	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.120	0.120	0.120	0.120	0.120	0.120	0.120	0.120	0.120
25	*M. darreli*	0.110	0.110	0.110	0.110	0.110	0.110	0.110	0.110	0.110	0.110	0.110	0.119	0.119	0.119	0.119	0.119	0.119	0.119	0.119	0.119
26	*M. chakrapanii*	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102
27	*M. ornata*	0.092	0.092	0.092	0.092	0.092	0.092	0.092	0.092	0.092	0.092	0.092	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099
28	*M. aurantiventris*	0.114	0.114	0.114	0.114	0.114	0.114	0.114	0.114	0.114	0.114	0.114	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109
29	*M. mihintalei*	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101
30	*M. nanapollexa*	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084
31	*M. pulchra*	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102
32	*M. berdmorei*	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.112	0.112	0.112	0.112	0.112	0.112	0.112	0.112	0.112
33	*M. heymonsi*	0.108	0.108	0.108	0.108	0.108	0.108	0.108	0.108	0.108	0.108	0.108	0.115	0.115	0.115	0.115	0.115	0.115	0.115	0.115	0.115
34	*M. orientalis*	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.122
35	*M. superciliaris*	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102

(Continued Table 2)

	Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
36	*M. petrigena*	0.063	0.063	0.063	0.063	0.063	0.063	0.063	0.063	0.063	0.063	0.063	0.077	0.077	0.077	0.077	0.077	0.077	0.077	0.077	0.077
37	*M. perparva*	0.058	0.058	0.058	0.058	0.058	0.058	0.058	0.058	0.058	0.058	0.058	0.072	0.072	0.072	0.072	0.072	0.072	0.072	0.072	0.072
38	*M. palmipes*	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.106	0.106	0.106	0.106	0.106	0.106	0.106	0.106	0.106
39	*M. mixtura*	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099
40	*M. butleri*	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107
41	*M. annectens*	0.079	0.079	0.079	0.079	0.079	0.079	0.079	0.079	0.079	0.079	0.079	0.072	0.072	0.072	0.072	0.072	0.072	0.072	0.072	0.072
42	*M. achatina*	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.103	0.103	0.103	0.103	0.103	0.103	0.103	0.103	0.103
43	*M. mantheyi*	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.105	0.105	0.105	0.105	0.105	0.105	0.105	0.105	0.105
44	*M. borneensis*	0.095	0.095	0.095	0.095	0.095	0.095	0.095	0.095	0.095	0.095	0.095	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.104	0.104
45	*M. malang*	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109
46	*M. mukhlesuri*	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099	0.099
47	*M. sholigari*	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.117	0.121	0.121	0.121	0.121	0.121	0.121	0.121	0.121	0.121
48	*M. mymensinghensis*	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.100	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102
49	*M. okinavensis*	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109
50	*M. rubra*	0.094	0.094	0.094	0.094	0.094	0.094	0.094	0.094	0.094	0.094	0.094	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101	0.101
51	*M. gadjahmadai*	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107
52	*M. laterite*	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.122	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.132	0.132
53	*M. beilunensis*	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102	0.102
54	*M. kodial*	0.113	0.113	0.113	0.113	0.113	0.113	0.113	0.113	0.113	0.113	0.113	0.115	0.115	0.115	0.115	0.115	0.115	0.115	0.115	0.115
55	*M. taraiensis*	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.109	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107
56	*M. nilphamariensis*	0.086	0.086	0.086	0.086	0.086	0.086	0.086	0.086	0.086	0.086	0.086	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084	0.084
57	*M. fissipes*	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.097	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107	0.107

	Species	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40
22	*M. marmorata*	0.082																			
23	*M. zeylanica*	0.135	0.131																		
24	*M. karunaratnei*	0.120	0.106	0.055																	
25	*M. darreli*	0.119	0.119	0.056	0.026																
26	*M. chakrapanii*	0.102	0.105	0.120	0.133	0.131															
27	*M. ornata*	0.099	0.103	0.124	0.104	0.099	0.100														
28	*M. aurantiventris*	0.109	0.124	0.117	0.094	0.099	0.132	0.106													
29	*M. mihintalei*	0.101	0.114	0.124	0.106	0.105	0.099	0.084	0.109												
30	*M. nanapollexa*	0.084	0.099	0.135	0.118	0.138	0.138	0.133	0.125	0.118											
31	*M. pulchra*	0.102	0.130	0.153	0.140	0.138	0.107	0.112	0.124	0.104	0.127										
32	*M. berdmorei*	0.112	0.115	0.126	0.120	0.113	0.120	0.102	0.120	0.100	0.105	0.104									
33	*M. heymonsi*	0.115	0.126	0.120	0.110	0.110	0.097	0.123	0.130	0.110	0.144	0.138	0.119								

(Continued Table 2)

	Species	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40
34	*M. orientalis*	0.122	0.118	0.138	0.130	0.131	0.091	0.128	0.135	0.104	0.111	0.121	0.092	0.085							
35	*M. superciliaris*	0.102	0.106	0.122	0.092	0.101	0.112	0.093	0.102	0.087	0.126	0.130	0.120	0.123	0.127						
36	*M. petrigena*	0.077	0.074	0.150	0.122	0.127	0.122	0.114	0.132	0.110	0.079	0.135	0.117	0.125	0.124	0.102					
37	*M. perparva*	0.072	0.065	0.151	0.128	0.130	0.120	0.119	0.119	0.120	0.089	0.133	0.122	0.124	0.138	0.110	0.053				
38	*M. palmipes*	0.106	0.108	0.145	0.112	0.117	0.106	0.086	0.108	0.094	0.127	0.117	0.126	0.125	0.132	0.094	0.099	0.099			
39	*M. mixtura*	0.099	0.104	0.112	0.120	0.118	0.062	0.109	0.111	0.106	0.127	0.111	0.110	0.089	0.087	0.111	0.127	0.115	0.118		
40	*M. butleri*	0.107	0.107	0.148	0.117	0.127	0.135	0.120	0.084	0.099	0.126	0.122	0.128	0.136	0.138	0.092	0.123	0.120	0.097	0.121	
41	*M. annectens*	0.072	0.072	0.164	0.143	0.148	0.136	0.132	0.135	0.133	0.106	0.155	0.123	0.148	0.150	0.125	0.082	0.079	0.129	0.146	0.136
42	*M. achatina*	0.103	0.123	0.138	0.140	0.136	0.092	0.108	0.128	0.102	0.122	0.096	0.085	0.089	0.063	0.128	0.128	0.136	0.124	0.084	0.125
43	*M. mantheyi*	0.105	0.122	0.130	0.135	0.134	0.079	0.125	0.130	0.099	0.125	0.121	0.098	0.087	0.058	0.117	0.132	0.138	0.129	0.082	0.138
44	*M. borneensis*	0.104	0.102	0.136	0.126	0.126	0.089	0.120	0.130	0.104	0.126	0.124	0.100	0.080	0.047	0.127	0.127	0.123	0.122	0.084	0.128
45	*M. malang*	0.109	0.110	0.133	0.123	0.129	0.087	0.115	0.128	0.104	0.121	0.117	0.105	0.082	0.051	0.120	0.127	0.128	0.117	0.089	0.120
46	*M. mukhlesuri*	0.099	0.109	0.132	0.123	0.126	0.044	0.090	0.132	0.099	0.130	0.106	0.112	0.089	0.081	0.097	0.114	0.120	0.101	0.060	0.117
47	*M. sholigari*	0.121	0.124	0.060	0.046	0.033	0.125	0.104	0.106	0.102	0.122	0.132	0.100	0.110	0.131	0.106	0.127	0.140	0.124	0.122	0.126
48	*M. mymensinghensis*	0.102	0.110	0.133	0.131	0.126	0.028	0.095	0.127	0.104	0.137	0.111	0.116	0.080	0.086	0.109	0.122	0.116	0.106	0.055	0.130
49	*M. okinavensis*	0.109	0.112	0.112	0.115	0.112	0.060	0.106	0.106	0.106	0.130	0.096	0.107	0.091	0.082	0.111	0.132	0.125	0.108	0.017	0.129
50	*M. rubra*	0.101	0.097	0.124	0.109	0.102	0.099	0.081	0.119	0.028	0.123	0.114	0.093	0.110	0.107	0.094	0.112	0.125	0.098	0.104	0.099
51	*M. gadjahmadai*	0.107	0.112	0.132	0.135	0.139	0.084	0.120	0.127	0.094	0.107	0.106	0.092	0.094	0.067	0.132	0.119	0.125	0.116	0.079	0.135
52	*M. laterite*	0.132	0.122	0.044	0.042	0.042	0.123	0.119	0.094	0.115	0.132	0.140	0.120	0.118	0.130	0.109	0.142	0.138	0.129	0.115	0.135
53	*M. beilunensis*	0.102	0.110	0.122	0.115	0.112	0.057	0.109	0.109	0.104	0.120	0.102	0.107	0.087	0.077	0.103	0.117	0.113	0.099	0.030	0.114
54	*M. kodial*	0.115	0.126	0.152	0.137	0.145	0.094	0.134	0.134	0.123	0.123	0.137	0.119	0.068	0.082	0.126	0.118	0.128	0.130	0.092	0.126
55	*M. taraiensis*	0.107	0.111	0.138	0.124	0.117	0.114	0.070	0.114	0.082	0.129	0.114	0.115	0.126	0.135	0.094	0.120	0.128	0.104	0.114	0.112
56	*M. nilphamariensis*	0.084	0.099	0.125	0.107	0.097	0.092	0.049	0.089	0.065	0.125	0.101	0.095	0.110	0.107	0.094	0.116	0.121	0.086	0.089	0.101
57	*M. fissipes*	0.107	0.107	0.115	0.123	0.121	0.033	0.097	0.130	0.096	0.130	0.109	0.107	0.090	0.072	0.097	0.114	0.118	0.092	0.055	0.125
	Species	**41**	**42**	**43**	**44**	**45**	**46**	**47**	**48**	**49**	**50**	**51**	**52**	**53**	**54**	**55**	**56**	**57**			
42	*M. achatina*	0.145																			
43	*M. mantheyi*	0.148	0.060																		
44	*M. borneensis*	0.138	0.067	0.060																	
45	*M. malang*	0.143	0.074	0.069	0.026																
46	*M. mukhlesuri*	0.141	0.077	0.077	0.086	0.074															
47	*M. sholigari*	0.143	0.136	0.125	0.131	0.134	0.127														
48	*M. mymensinghensis*	0.142	0.077	0.079	0.084	0.077	0.030	0.123													
49	*M. okinavensis*	0.152	0.087	0.086	0.087	0.087	0.055	0.117	0.057												
50	*M. rubra*	0.125	0.105	0.107	0.100	0.102	0.094	0.097	0.099	0.149											
51	*M. gadjahmadai*	0.152	0.058	0.058	0.067	0.076	0.081	0.128	0.083	0.143	0.097										

(Continued Table 2)

	Species	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57
52	*M. laterite*	0.162	0.135	0.128	0.133	0.133	0.130	0.046	0.126	0.151	0.114	0.125						
53	*M. beilunensis*	0.144	0.078	0.077	0.082	0.087	0.058	0.117	0.055	0.130	0.102	0.072	0.114					
54	*M. kodial*	0.144	0.082	0.077	0.080	0.089	0.072	0.139	0.079	0.135	0.123	0.084	0.147	0.092				
55	*M. taraiensis*	0.141	0.131	0.117	0.120	0.127	0.104	0.110	0.107	0.135	0.074	0.120	0.135	0.113	0.126			
56	*M. nilphamariensis*	0.127	0.093	0.105	0.099	0.102	0.084	0.097	0.087	0.130	0.062	0.092	0.112	0.092	0.115	0.055		
57	*M. fissipes*	0.134	0.080	0.075	0.082	0.077	0.026	0.118	0.033	0.138	0.092	0.077	0.118	0.049	0.075	0.101	0.089	

well beyond snout tip (Table 4).

The new form of *Microhyla* from Lam Dong Province most closely resembles *M. pulchella* in morphology as well as supported by molecular phylogenetic relationships (Figure 2), but they are differentiated from each other in the following characteristics: The new form has a notably slender body habitus than that of *M. pulchella*, which is reflected in a number of diagnostic morphometric characteristic ratios to SVL (Figures 4, 7E, Table 6). Males of *M. pulchella* have two small black scapular spots on dorsum, both usually bordered by a thin white line (*vs.* absent in males of the new form, see Figures 4, 7E, 7F) (Poyarkov *et al.*, 2014). Dorsum skin of the new form scattered by small tubercles (*vs.* dorsum skin smooth in *M. pulchella*). *M. pulchella* have few black scapular spots behind the eyes (*vs.* only one small black spot behind the eye in the new form, see Figures 4, 9) (Poyarkov *et al.*, 2014). *M. pulchella* has a median longitudinal groove on dorsal surface of finger disc (*vs.* absent in the new form). *M. pulchella* also differs from the new form in the webbing formula (I1–2II1–2½III1–2½IV2½–1V *vs.* I1½–2II1– 2III1–2½IV2¼–1V in the new form) (Table 6).

Moreover, statistical results indicated that the new form of *Microhyla* from Lam Dong Province could be separated from *M. pulchella* based on morphometric data. PCA extracted four principal component axes with eigenvalues greater than 0.001, and of these, the first two component axes accounted for 82.19% of the variation (Table 5). The first two principal component axes could separate the new form from *M. pulchella* by 19 characteristics (Figures 6A, 6B), mainly based on limb and head measurements, namely: HL, SL, EL, NEL, HW, IND, UEW, FLL, LAL, HAL, 1FL, OPTL, 3FDD, HLL, TL, FL, IMTL, 1TOEL, 3TDD (Table 5). Species with a larger and positive score on PC1 reflected shorter SVL, HL, SL, EL, NEL, HW, IND, UEW, FLL, LAL, HAL, 1FL, OPTL, 3FDD, HLL, TL, FL, IMTL, 1TOEL and 3TDD while a negative score signified smaller IOD. The PC2 with negative scores was associated with species having shorter SL, EL, NEL, IND, 1TOEL, 3TDW whereas positive scores were associated with species with larger morphological traits such as HL, HW, IOD, UEW, FLL, LAL, HAL, OPTL, 1FL, 3FDW, HLL, TL and FL (Table 5).

3.3. Description

***Microhyla hongiaoensis* sp. nov.**

(Figures 3A, 3B, 3C, 3D, 3E, 3F; Tables 3, 4)

Holotype VNMN 07477, adult male, collected in Hon Giao Forest, Bi Doup-Nui Ba National Park, Da Chai Commune, Lac Duong District, Lam Dong Province (12°11'18.3"N, 108°40'29.0"E, elevation ca. 1548 m a.s.l., Figure 1); collected by C.V. Hoang *et al.* on 8 May 2018.

Paratypes (n = 10) All specimens collected by C.V. Hoang *et al.*, the same location data as the holotype: 2 adult males (VNMN

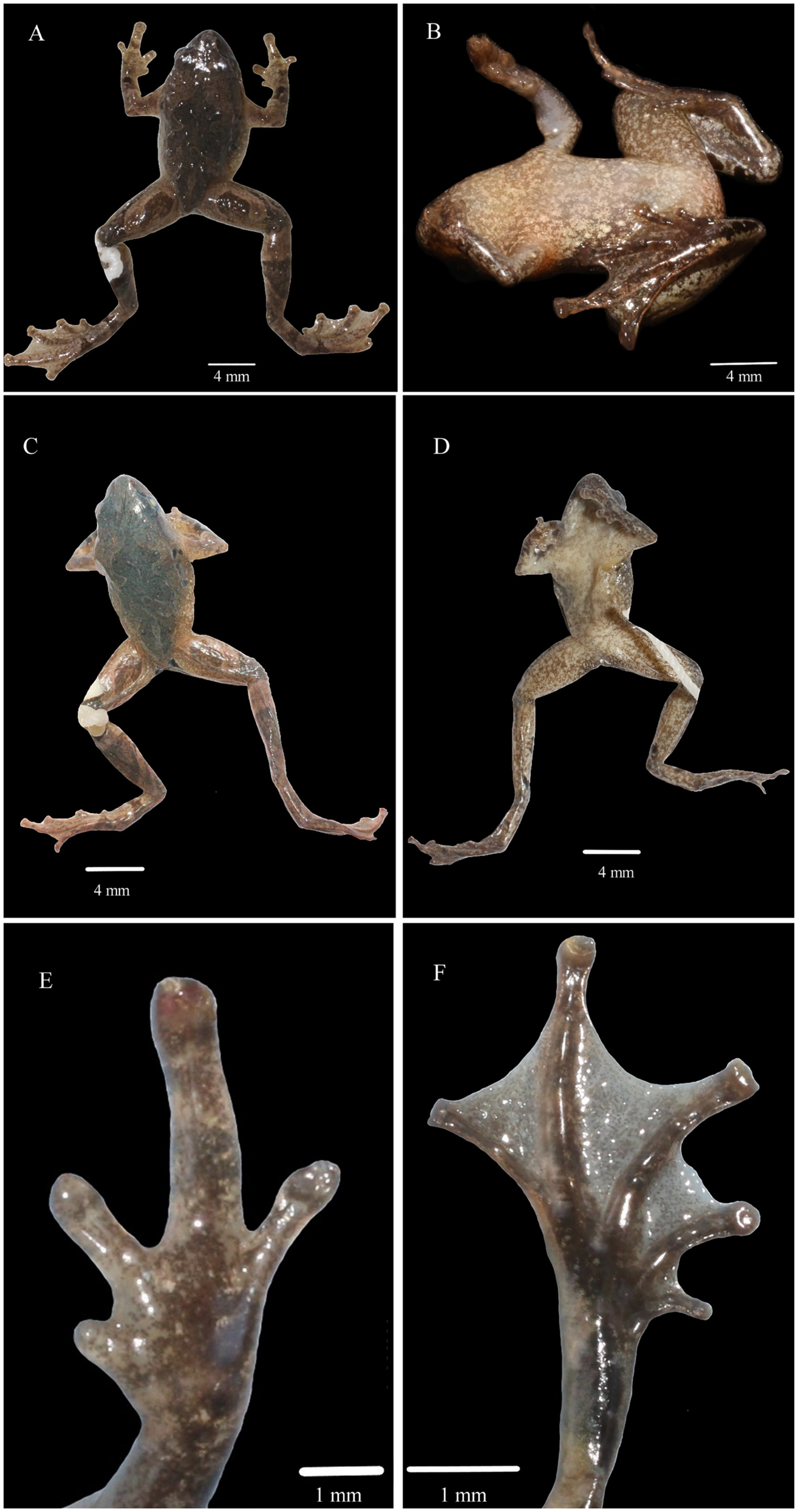

Figure 3 Dorsal and ventral views of the holotype (VNMN07477, male) of *Microhyla hongiaoensis* **sp. nov.**: in life (A, B) and in preservative (C, D); underside of right hand (E) and right foot (F) . Photos by Chung V. Hoang.

Table 3 Measurements (in mm) and proportions of the type series of *Microhyla hongiaoensis* **sp. nov.** and *M. pulchella*.

Species	*M. hongiaoensis* **sp. nov.**				*M. pulchella*			
Sex	Female (*n* = 3)		Male (*n* = 8)		Female (*n* = 4)		Male (*n* = 17)	
	Mean ± SD	Min–Max	Mean ± SD	Min–Max	Mean ± SD	Min–Max	Mean ± SD	Min–Max
SVL	18.43 ± 0.18	18.28–18.63	14.1 ± 0.39	13.55–14.65	23.39 ± 1.63	21.77–25.4	17.43 ± 0.51	16.53–18.22
HL	5.65 ± 0.16	5.54–5.83	4.39 ± 0.29	3.92–4.72	5.87 ± 0.2	5.66–6.13	4.89 ± 0.3	4.32–5.4
HW	5.36 ± 0.21	5.13–5.53	4.23 ± 0.29	3.82–4.88	6.24 ± 0.36	5.87–6.56	5.34 ± 0.27	5.02–5.83
SL	2.64 ± 0.06	2.58–2.69	1.93 ± 0.06	1.84–2.01	2.9 ± 0.13	2.82–3.1	2.4 ± 0.15	2.11–2.59
EL	1.89 ± 0.1	1.77–1.96	1.44 ± 0.1	1.3–1.57	2.18 ± 0.22	1.88–2.41	1.88 ± 0.17	1.57–2.31
N-EL	1.52 ± 0.21	1.28–1.66	1.08 ± 0.11	0.94–1.3	1.56 ± 0.26	1.25–1.79	1.34 ± 0.06	1.22–1.44
IND	1.31 ± 0.15	1.14–1.43	0.97 ± 0.13	0.83–1.25	1.79 ± 0.16	1.56–1.94	1.5 ± 0.11	1.27–1.68
IOD	2.72 ± 0.04	2.69–2.76	2.39 ± 0.17	2.13–2.65	3.15 ± 0.48	2.58–3.65	2.35 ± 0.18	1.97–2.74
UEW	0.9 ± 0.06	0.84–0.95	0.72 ± 0.08	0.62–0.87	1.37 ± 0.25	1.07–1.67	1.05 ± 0.1	0.92–1.29
FLL	11.02 ± 0.12	10.89–11.13	9.51 ± 0.37	9.13–10.28	14.31 ± 1.1	13.54–15.93	13.48 ± 0.65	12.02–14.51
LAL	8.5 ± 0.31	8.3–8.85	7.5 ± 0.36	6.81–7.89	10.75 ± 0.95	10.07–12.15	10.32 ± 0.45	9.5–11.14
HAL	4.64 ± 0.22	4.43–4.87	4.44 ± 0.26	4.09–4.83	5.73 ± 0.71	5.15–6.75	5.82 ± 0.36	5.09–6.27
IPTL					0.92 ± 0.1	0.83–1.04	0.68 ± 0.08	0.61–0.87
OPTL	0.49 ± 0.02	0.47–0.51	0.41 ± 0.05	0.36–0.52	1.06 ± 0.05	1.02–1.13	0.76 ± 0.09	0.68–0.96
1FL	1.38 ± 0.1	1.32–1.49	0.91 ± 0.25	0.43–1.25	1.61 ± 0.09	1.53–1.7	1.59 ± 0.22	1.3–1.99
1FW	0.37 ± 0.09	0.3–0.47	0.27 ± 0.04	0.21–0.33	0.4 ± 0.06	0.34–0.46	0.35 ± 0.07	0.21–0.47
1 FLO	0.62 ± 0.15	0.48–0.78	0.31 ± 0.06	0.22–0.41	0.85 ± 0.19	0.72–1.13	0.62 ± 0.19	0.32–0.97
2 FLO	1.19 ± 0.23	0.97–1.43	1.21 ± 0.17	0.92–1.37	1.52 ± 0.22	1.25–1.76	1.47 ± 0.19	1.15–1.79
3 FLO	2.6 ± 0.1	2.51–2.71	2.59 ± 0.29	2.18–2.95	3.58 ± 0.27	3.39–3.96	3.73 ± 0.12	3.5–4.04
2 FLI	1.76 ± 0.06	1.72–1.83	1.54 ± 0.18	1.21–1.76	2.37 ± 0.45	2.09–3.04	2.08 ± 0.27	1.77–2.45
3 FLI	2.66 ± 0.15	2.49–2.77	2.59 ± 0.26	2.15–2.86	3.96 ± 0.2	3.75–4.17	4.09 ± 0.32	3.39–4.51
4 FLI	1.27 ± 0.15	1.13–1.43	1.01 ± 0.22	0.74–1.4	1.59 ± 0.17	1.36–1.77	1.33 ± 0.24	0.77–1.75
2 FDW	0.36 ± 0.08	0.31–0.45	0.45 ± 0.08	0.33–0.55	0.46 ± 0.05	0.38–0.5	0.52 ± 0.08	0.4–0.64
3 FDW	0.44 ± 0.02	0.43–0.46	0.6 ± 0.1	0.45–0.79	0.53 ± 0.13	0.38–0.69	0.74 ± 0.14	0.38–0.88
4 FDW	0.38 ± 0.07	0.34–0.46	0.33 ± 0.04	0.28–0.4	0.46 ± 0.07	0.36–0.53	0.37 ± 0.06	0.25–0.51
HLL	33.86 ± 0.73	33.29–34.68	26.45 ± 0.88	25.04–27.68	44 ± 4.52	41.02–50.73	34.67 ± 1.33	32.67–36.59
TL	11.1 ± 0.24	10.94–11.38	8.68 ± 0.11	8.52–8.84	14.42 ± 1.36	13.26–16.36	11.32 ± 0.44	10.5–12.04
FL	14.6 ± 1	13.48–15.41	11.84 ± 0.17	11.51–12.06	18.03 ± 1.41	17.18–20.12	14.45 ± 0.79	13.4–16.51
1TOEL	1.26 ± 0.47	0.9–1.79	0.87 ± 0.18	0.63–1.09	3.31 ± 0.46	2.73–3.79	2.3 ± 0.43	1.38–2.88
2TOEL	2.39 ± 0.21	2.18–2.6	1.86 ± 0.29	1.36–2.27	4.44 ± 0.61	3.62–5.1	3.49 ± 0.34	3.02–3.98
3TOEL	3.91 ± 0.58	3.26–4.37	3.41 ± 0.49	2.42–3.92	6.74 ± 0.75	5.94–7.75	5.16 ± 0.37	4.6–5.85
4TOEL	6.47 ± 0.6	5.8–6.97	4.78 ± 0.66	3.78–5.61	8.59 ± 1.8	7.13–11.19	6.73 ± 0.41	6.19–7.67
5TOEL	4.3 ± 0.35	3.92–4.61	2.86 ± 0.33	2.32–3.52	4.89 ± 0.83	3.96–5.95	3.77 ± 0.29	3.09–4.22
IMTL	0.6 ± 0.04	0.55–0.63	0.5 ± 0.16	0.28–0.71	0.83 ± 0.17	0.71–1.08	0.53 ± 0.11	0.24–0.67
1 TDW	0.54 ± 0.1	0.45–0.65	0.37 ± 0.06	0.31–0.49	0.69 ± 0.04	0.65–0.75	0.49 ± 0.08	0.35–0.64
2 TDW	0.9 ± 0.06	0.85–0.96	0.56 ± 0.09	0.42–0.71	1.14 ± 0.22	0.87–1.4	0.79 ± 0.08	0.69–0.95
3 TDW	1.11 ± 0.13	0.99–1.24	0.69 ± 0.1	0.57–0.8	1.28 ± 0.16	1.05–1.42	0.95 ± 0.1	0.77–1.17
4 TDW	0.98 ± 0.19	0.77–1.15	0.59 ± 0.09	0.47–0.7	1.39 ± 0.14	1.22–1.55	0.91 ± 0.07	0.76–1.02
5 TDW	0.72 ± 0.08	0.64–0.8	0.52 ± 0.11	0.32–0.67	1.13 ± 0.12	1.01–1.29	0.63 ± 0.15	0.34–0.97

07385, 07388) and 1 adult gravid female (VNMN 07390), collected on 7 May 2018; 5 adult males (IEBR 4573 = VNMN 07405, IEBR 4574 = VNMN 07406, IEBR 4575 = VNMN 07718, VNMN 07483, VNMN 07617) and 2 adult gravid females (IEBR 4576 =VNMN 07478, VNMN 07448), collected on 8 May 2018.

Diagnosis *Microhyla hongiaoensis* **sp. nov.** is assigned to the genus *Microhyla* based on the molecular phylogenetic data and the following morphological characteristics: relatively small body size; maxillary and vomerine teeth absent; vomer divided into two parts, disappearing at the posterior edge of the choana; tongue round posteriorly; skin smooth or with tubercles; tympanum hidden; palate with 1–2 rows of horizontal skin ridges; fingers without webbing; toes slightly webbed or free of webbing; metacarpal tubercles 2 or 3; and the absence of skin

Table 4 Selected diagnostic characters for the species in the genus *Microhyla* (modified from Poyakov *et al*., 2014; Nguyen *et al*., 2019). All measurements were given in millimeter (mm).
Note: F1, relative length of finger I (I < one-half length of II, I > one-half length of II, present as a nub or pronounced bulge); FD, disks on distal end of fingers; FMG, dorsal median longitudinal grooves on finger disks; TD, disks on distal end of toes; TMG, dorsal peripheral grooves on toe disks; MTT, number of metatarsal tubercles; DML, presence of absence of light dorsomedial (vertebral) line; SCT, presence or absence of superciliary tubercles; Presence: √; Absent: ×; Tibiotarsal, where on body tibiotarsal projection stretches at adpressed limb; Webbing, webbing formula according to Savage (1975), if it was not possible, extent of webbing on feet is described in words. For references see below. ** It is unknown whether the only know specimen of *M. fusca* is male or female;*** Measurements of F1 in Bain and Nguyen (2004) are different from those of Matsui (2011) used in the present paper.

	Species	SVL		Habitus	Snout profile	Dorsum skin	F1***	FD	FMG	TD	TMG	MTT	SCT	DML	Tibiotarsal	Foot webbing	Distribution
		Male	Female														
	M hongiaoensis **sp. nov.**	13.6–14.7	18.3–18.6	Slender	Bluntly rounded	Scattered by small tubercles	F1 < ½ F2	√, weak F2–F4	×	√	√, weak T2-T5	2	×	×	Well beyond snout	I1–2II1–2½III1–2½IV2½–1V	Langbian pl. S Vietnam
1	*M. achatina*	16	23	Slender	Obtusely pointed	Smooth or feebly tubercular	F1 < ½ F2	√	√	√	√	2	×	√	To snout or just beyond	I2–2½II2–3¼III3–4IV 4–3V	Java, Bali
2	*M. annamensis*	15.2–19.8	18.2–22.6	Moderately stocky	Bluntly rounded	Warty, strongly tubercular	F1 < ½ F2	√	√	√	√	2	×	×	Well beyond snout	I1–2¼II1–2½III1½–2¾IV3–1V	Langbian pl. S Vietnam
3	*M. annectens*	14.4–15.6	18.2–18.4	Slender	Rounded	Smooth	F1 < ½ F2	√	√	√	√	1	×	×	Well beyond orbit	I1–1II1–1III1–3IV3–1V	Malaya, Borneo, Philippines
4	*M. arboricola*	13.2–15.0	15.9–17.0	Moderately slender	Pointed	Feebly granular	F1 < ½ F2	√, F2–F4	√, weak	√	√	1	×	×	Well beyond snout	I1⅔–2¼II2–3III2½–3½IV3–1½V	Langbian pl., S Vietnam
5	*M. aurantiventris*	25.2–27.0	30	Moderately stocky	Rounded	Slightly shagreened with numerous tiny tubercles	F1 > ½ F2	√, F2–F4	√, weak	√	√	2	×	×	Reaching slightly beyond snout	I1¾–2II1½–2¾III2–3⅓IV3¼–1½V	Kon Tum pl., S Vietnam
6	*M. beilunensis*	19.1–23.7	26.1–28.3	Moderately slender	Bluntly rounded	Smooth, small tubercles	F1 < ½ F2	√, weak	×	√	√	2	×	×	To the eye	Basal	Zhejiang Province, China
7	*M. berdmorei*	23.8–28.9	26.2–45.6	Slender	Obtusely pointed	Smooth, small tubercles	F1 < ½ F2	√, weak	√	√	√	2	×	×	Well beyond snout	I1–1II1–2III1–2IV2–1V	S China, SE Asia to G. Sundas
8	*M. borneensis*	10.6–12.8	17.9–18.8	Stocky	Obtusely pointed	Smooth, small tubercles	nub or bulge	weak or ×	√	√	√	2	×	×	Shorter than snout	I1–2II1–3III2½–3⅓IV3½–2V	Sarawak
9	*M. butleri*	20.0–25.0	21.0–26.0	Slender	Rounded	Smooth or tubercular	F1 > ½ F2	√, weak	√	√	√	2	×	×	Shorter than snout	I2–2½II1¾–3III2⅓–3½IV3½–2¼V	S China, SE Asia to Malaya
10	*M. chakrapanii*	22	?	Moderately stout	Obtusely rounded	Smooth	F1 > ½ F2	×	×	√	×	2	×	×	Beyond snout (?)	Basal; between T3–T4 to proximal tubercle	Andamans
11	*M. darevskii*	27.0–32.6	?	Stocky, flattened	Rounded	Slightly tubercular or pustulate	F1 > ½ F2	×	×	weak	√	2	×	×	Well beyond snout	I1–1II1–1III1–1IV1–1V	Ngoc Linh mt., C Vietnam
12	*M. darreli*	15.0–15.7	?	Rather slender	Subovoid	Shagreened to sparsely granular	F1 > ½ F2	√, weak	×	√, weak	√	2	×	×	Shorter than eye	I2–2¼II1¾–3III2–3IV3–2¼V	S India
13	*M. fanjingshanensis*	19.0–22.7	22.5–23.0	Slender	Rounded	Roughish with tiny tubercles	F1 > ½ F2	×	×	√	√	2	×	√	Between eye to nostril	Basal	Guizhou Province, China
14	*M. fissipes*	18.0–27.5	20.0–28.0	Moderately slender	Rounded	Smooth or slightly tubercular	F1 > ½ F2	×	×	×	×	2	×	×	Shorter than snout	I2–2½II2–3½III3–4IV4–3V	S Thailand, Indochina to Malaya
15	*M. fodiens*	12.6–20.8	?	Stout	Rounded	Feebly tubercular	F1 < ½ F2	×	×	×	×	2	×	×	Not reaching orbit level	I1–2II1¾–3III2¾–3¾IV4–2¾V	Myanmar
16	*M. fowleri*	29.5–32.5	32.2–41.5	Stocky	Obtusely pointed	Rugose, pustular	F1 > ½ F2	weak or ×	×	√	√	2	×	×	Well beyond snout	I1–1II1–1III1–2IV2–1V	N Thailand, S China
17	*M. fusca*	23.0**		Slender	Acuminate	Shagreened, faint middorsal ridge	F1 > ½ F2	√, F3	?	weak, T2–T5	√, T4	2	×	×	To the eye	Basal, continue as folds up toes	Langbian pl., S Vietnam
18	*M. gadjahmadai*	18.20–21.32	20.37–25.51	Stout body	Dorsally rounded	Low tubercles	F1 > ½ F2	√, weakly dilated	√	√	√	2	×	√	Well beyond snout	I2–2¼II1¾–3III3–4IV4–2¾V	Sumatra, Indonesia

(Continued Table 4)

	Species	SVL		Habitus	Snout profile	Dorsum skin	FI***	FD	FMG	TD	TMG	MTT	SCT	DML	Tibiotarsal	Foot webbing	Distribution
		Male	Female														
19	*M. heymonsi*	16.5–22.0	18.0–26.5	Stocky	Rounded	Smooth	FI ≤ ½ F2	√	usually √	√	Usually √	2	×	√	Shorter than snout	I2–2⅓II2–3III3–4IV4⅓–3V	S China, NE India, SE Asia to Sumatra
20	*M. irrawaddy*	12.3–17.1	16.7–20.9	Very slender	Acuminate	Granular	FI > ½ F2	√, F2–F4	×	×	√, weak T2-T5	?	×	×	Reaching eye level	I2–3II2–3III3–4½IV4½–2¾V	Myanmar
21	*M. karunaratnei*	15.8–19.1	19.6–21.0	Moderately stocky	Rounded	Smooth	FI > ½ F2	√	√	√	√	2	×	×	Beyond snout (?)	I2–2½II2–3½III2½–3¾IV4–2V	S Sri Lanka
22	*M. kodial*	16.9–17.4	18.0–20.4	Slender	Rounded	Tuberculated	FI > ½ F2	√, F2–F4	×	√	×	2	×	×	Well beyond snout	weak, Basal	W S India
23	*M. laterite*	15.3–16.6	18.4	Very small sized	Obtuse	Smooth	FI > ½ F2	√	√	√	√	2	√	×	Well beyond snout	I1–2II1–2III1⅔–2IV3–1V	W S India
24	*M. maculifera*	12.0–13.3	11.8	Moderately stout	Bluntly rounded	Dorsolateral rows of tubercles	FI > ½ F2	×	×	√, weak	×	1	×	×	To snout or just beyond	Basal	Sabah, Borneo
25	*M. malang*	18.7–22.2	19.0–23.4	Stocky	Rounded	Smooth	FI < ½ F2	√	√	√	√	2	×	×	To snout or just beyond	I1–2II1–2⅔III1⅔–3IV3–1V	Borneo
26	*M. mantheyi*	15.0–29.2	14.8–24.1	Stocky	Pointed	Granular, feebly pustular	FI < ½ F2	√	√	√	√	2	×	×	Well beyond snout	I1–2II1–2III2–3IV3–1½V	S Thailand, Malaya
27	*M. marmorata*	18.8–21.5	21.1–23.2	Moderately stocky	Bluntly rounded	Smooth or feebly pustular	FI < ½ F2	√	√	√	√	2	×	×	Well beyond snout	I1–2II1–1¾III1½–2¾IV2¾–1V	C Vietnam, Laos
28	*M. mihintalei*	21.7–27.3	24.4	Slender	Sub-ovoid	Smooth	FI ≤ ½ F2	×	×	×	×	2	√	×	To snout	basal	Sri Lanka
29	*M. minuta*	14.7–15.9	15.7–17.2	Slender	Bluntly rounded	Granular, feebly pustular	FI ≤ ½ F2	√, F2–F4	√, weak	√	√	2	×	×	Shorter than snout, but beyond the eye level	In.a.– n.a.II2–3½III3–4IV4–3V	Dong Nai, S Vietnam
30	*M. mixtura*	20.5–23.7	23.8–26.6	Stout	Rounded	Smooth, with tubercles	FI < ½ F2	√, weak	×	√	√	2	×	×	Shorter than snout	I2–2½III¾–3¼III3–4IV4¼–2¾V	C & E China
31	*M. mukhlesuri*	16.5–21.0	17.3–18.4	Moderately slender	Rounded	Smooth	FI > ½ F2	×	×	×	×	2	×	×	To snout	I2–2½II2–3½III3–4IV4–2¾V	Bangladesh
32	*M. mymensinghensis*	14.2–17.6	15.2–21.3	Stocky	Truncated	Smooth	FI > ½ F2	×	×	×	×	2	×	×	To snout	I2–2½II2–3½III3–4IV4¼–2¾V	Bangladesh
33	*M. nanapollexa*	?	16.6	Slender	Rounded	Smooth	nub or bulge	√	√	√	√	1	×	×	Well beyond snout	I1–2II1–2½III2½–2½IV 2½–1V	Ngoc Linh mt., C Vietnam
34	*M. nilphamariensis*	14.8–20.0	18.7–21.0	Stout	Rounded	Smooth	FI > ½ F2	×	×	×	×	2	×	×	To snout	Basal	Bangladesh, central and eastern Nepal, N India
35	*M. okinavensis*	22.5–28.2	26.5–30.8	Moderately slender	Rounded	Smooth or slightly tubercular	FI ≤ ½ F2	×	×	×	×	2	×	×	To snout	I1½–2II1½–3¼III2¾–4IV4–2½V	Okinawa
36	*M. orientalis*	15.8–17.4	15.8–17.4	Moderately slender	Rounded	Smooth or slightly tubercular	FI < ½ F2	√, weak F2–F4	√	√	√	2	×	√	To the eye	In.a.– n.a.II2–3¼III3–4¼IV4¼–3V	Bali
37	*M. ornata*	13.4– 24.9	24.9–26.2	Moderately slender	Rounded	Smooth or slightly tubercular	FI ≤ ½ F2	×	×	×	×	2	×	×	Shorter than snout	I2–2½III¾–3½III3–4IV4–2¾V	Sri Lanka, India to Andamans
38	*M. palmipes*	16	21.8	Slender	Rounded	Smooth or slightly tubercular	nub or bulge	√	×	√	×	2	×	×	To snout or just beyond	In.a.– n.a.II2⅓–3¾III3¼–4IV4–3V	Malaya & Sundas
39	*M. perparva*	10.5–11.9	12.4–14.5	Moderate	Obtusely pointed	Smooth	nub or bulge	√	×	√	√	1	√	×	Well beyond snout	I1–1II1–1III1–2IV2–1V	Borneo
40	*M. petrigena*	13.9–16.2	15.1–17.8	Moderately stout	Obtusely pointed	Smooth, flank and posterior tubercles	nub or bulge	√	√, weak	√	√	1	×	×	Well beyond snout	I1–1II1–1III1–2IV2–1V	Borneo

(Continued Table 4)

Species	SVL Male	SVL Female	Habitus	Snout profile	Dorsum skin	F1***	FD	FMG	TD	TMG	MTT	SCT	DML	Tibiotarsal	Foot webbing	Distribution
41 *M. picta*	25.2–30.1	27.2–33.4	Stout	Rounded	Smooth or slightly warty	F1 < ½ F2	×	×	×	×	2	×	×	Shorter than eye	I2–2¾II1¾–2¾III2¾–3¾IV4–2½V	SE Vietnam
42 *M. pineticola*	17.2–19.5	18.0–23.0	Stocky	Acuminate	Smooth	F1 < ½ F2	√, F2–F4	√	√	√, weak	2	×	√	Shorter than snout, but beyond the eye level	I1½–2½II1¾–3III2¾–3¾IV4–2½V	Langbian pl., S Vietnam
43 *M. pulchella*	14.7–21.6	18.1–25.8	Moderately stocky	Bluntly rounded	Smooth	F1 < ½ F2	√, F2–F4	√, weak	√	√, weak	1(2)	×	×	Well beyond snout	I1½–2II1–2III1–2½IV2¼–1V	Langbian pl., S Vietnam
44 *M. pulchra*	23.0–32.0	28.0–36.5	Stocky	Obtusely pointed	Smooth, feebly granular	F1 < ½ F2	×	×	×	×	2	×	×	To snout or just beyond	I1½–2II1–3III2–3¼IV3½–2V	S China, Thailand, Indochina
45 *M. pulverata*	17.5–19.5	18.8–20.2	Moderately stocky	Bluntly rounded	Smooth or feebly pustular	F1 < ½ F2	√	√	√	√	2	×	×	Well beyond snout	I1–2II1–2½III1¾–3IV3–1V	C Vietnam
46 *M. rubra*	20.0–27.5	20.5–29.5	Stout	Rounded	Smooth, feebly tuberculated	F1 ≤ ½ F2	×	×	×	×	2	×	×	Shorter than snout	I1½–2II1½–3III2½–3IV4–2½V	S & E India, Sri Lanka
47 *M. sholigari*	?	11.0–15.0	Moderately slender	Truncated	Smooth	F1 > ½ F2	√	×	√	√	2	×	√	Shorter than snout	I1½–2II2½–3½III2½–3½IV3¾–2V	SW India
48 *M. superciliaris*	?	12	Slender	Rounded	Smooth	F1 < ½ F2	√	×	√	√, weak	2	√	×	To snout or just beyond	I1–1II1–1III1–2IV2–1V	Malaya, Sumatra
49 *M. taraiensis*	19.9–20.9	22.1–24.9	Stout	Rounded	granular	F1 > ½ F2	×	×	×	×	2	×	×	To snout	I2–3II2¾–3¾III3–4¼IV4–2¾V	E Nepal
50 *M. zeylanica*	14.4–18.3	15.8–20.0	Moderately slender	Rounded	Smooth or slightly tubercular	F1 > ½ F2	×	×	√	×	2	×	×	To the eye	I2–2¼II1¾–3½III2¼–3¾IV4–2V	C Sri Lanka

ridge or skin projection between the subarticular tubercles of toes III and IV.

Microhyla hongiaoensis **sp. nov.** is distinguished from its congeners by a combination of the following characteristics: (1) size medium (SVL 13.6–14.7 mm in males and 18.3–18.6 mm in females); (2) fingers II–IV with small disks, dorsal surface of disks, without median longitudinal groove; (3) webbing formula I1½ – 2II1 – 2III1 – 2½IV2¼ – 1V; (4) dorsal surface of toe disks with median longitudinal groove; (5) dorsal back without two small black spots; (6) one small black spot present behind the eye; (7) few small black scapular spots in the flank-belly and inguinal region; (8) palm with two small metatarsal tubercles; (9) tibiotarsal reaching beyond snout.

Description of holotype Small-sized frog, SVL 14.3 mm; habitus slender, head longer than wide (HL/HW 1.1); snout bluntly round, slightly protruding beyond margin of lower jaw, longer than diameter of eye (SL/EL 1.41); eyes protuberant, pupil round; dorsal surface of head flat, canthus rostralis round, distinct; loreal region oblique, weakly concave; nostril round, lateral, on canthus rostralis, closer to tip of snout than to eye (NEL 1.3 mm); interorbital distance wide (IOD 2.3 mm), much greater than internarial distance (IND 0.9 mm) and upper eyelid width (UEW 0.8 mm); tympanum hidden, supratympanic fold weakly developed, from posterior corner of eye to arm insertion; vomerine teeth absent, tongue round posteriorly and free at the rear half of its length; slit-like openings to a median vocal sac.

Forelimbs relatively short, about three times shorter than hindlimbs (FLL/HLL 0.4); hand shorter than a half of forelimb length (HAL/FLL 0.5); fingers slender, free of webbing, dorsoventrally flattened, fingers without skin fringes; the first finger slightly reduced notably, less than one-half the length of the second finger (1FLO/2FLO 0.2); the second finger slightly longer than fourth (2FLI/4FLI 1.7), and much shorter than the third (2FLI/3FLI 0.6); relative finger lengths: I<IV<II<III. Tip of first finger round, not enlarged; tips of fingers II–IV slightly dilated, forming weak round disks; same width as basal phalanges on the fourth and second fingers, much narrower than basal phalange on the third finger; third finger with basal phalange twice larger than that of the first finger (3FDW/1FW 2.0); narrow peripheral grooves absent on all fingers; grooves on dorsal surface of disks present in fingers II, III, IV, absent in the first finger; relative finger disk widths: IV<II<III; subarticular tubercles on fingers distinct, round, formula: 1 : 1 : 2 : 1 (given for fingers I : II : III : IV, respectively); thumb present on anterior part of forearm; nuptial pad absent; inner palmar tubercle absent, outer palmar tubercle round and prominent.

Hindlimbs slender and long (HLL 26.2), tibia longer than half of snout-vent length (TL/SVL 0.6); tibiotarsal articulation reaching beyond snout when limb adpressed along body; foot longer than tibia (FL/TL 1.3); relative toe lengths: I<II<V<III<IV;

Table 5 Variable loadings for principal components with eigenvalue greater than 0.01, from morphometric characters corrected by SVL. All measurements were given in millimeter (mm).

Male	Axis 1	Axis 2	Axis 3	Axis 4	Axis 5
SVL	0.13	0	0.15	0.04	0.08
HL	0.08	−0.03	0.21	0	0.25
HW	0.14	−0.03	0.25	0.08	0.3
SL	0.14	0.01	0.15	0.05	0.16
EL	0.16	0.06	0.08	0.05	0.09
N-EL	0.14	0.02	0.11	−0.06	0.2
IND	0.26	0.07	0.13	0.08	0.03
IOD	−0.01	−0.02	0.13	0.02	0.13
UEW	0.24	−0.04	0.11	−0.01	0.27
FLL	0.19	−0.1	0.11	−0.01	−0.08
LAL	0.18	−0.1	0.05	0	−0.05
HAL	0.16	−0.05	0.02	−0.11	−0.02
OPTL	0.37	−0.03	0.16	0.13	0.03
1FL	0.31	−0.53	−0.39	−0.59	0.18
3_FDW	0.11	−0.17	−0.54	0.68	0.38
HLL	0.16	−0.03	0.08	0	0
TL	0.15	−0.02	0.12	−0.03	0.03
FL	0.1	−0.07	−0.05	−0.07	−0.21
1TOEL	0.57	0.26	−0.28	0.13	−0.54
IMTL	0.11	0.76	−0.28	−0.33	0.38
3_TDW	0.2	0.04	0.34	0.07	−0.04
Eigenvalue	0.13	0.02	0.01	0.01	0.01
% variance	71.59	10.6	4.77	4.01	3.71
Female	Axis 1	Axis 2	Axis 3	Axis 4	Axis 5
SVL	0.15	0.12	0.11	−0.03	−0.05
HL	0.10	−0.04	−0.06	0.02	−0.08
HW	0.10	0.12	0.02	−0.06	−0.23
SL	0.06	−0.01	−0.07	−0.04	0.19
EL	0.09	0.18	−0.17	0.13	0.33
N-EL	0.05	−0.20	−0.16	0.67	0.44
IND	0.20	0.16	−0.15	0.16	0.02
IOD	0.10	0.33	0.14	0.21	−0.25
UEW	0.26	−0.33	0.23	0.07	−0.25
FLL	0.16	−0.01	0.14	0.01	−0.07
LAL	0.15	0.04	0.15	0.13	0.10
HAL	0.13	−0.03	0.29	0.15	−0.12
OPTL	0.45	0.04	0.18	−0.47	0.55
1FL	0.09	0.11	0.18	−0.16	−0.10
3_FDW	0.10	0.66	−0.21	−0.07	0.04
HLL	0.17	0.05	0.18	0.11	−0.02
TL	0.17	0.04	0.08	0.12	−0.03
FL	0.13	0.01	0.26	0.04	0.17
1TOEL	0.64	−0.29	−0.51	−0.06	−0.25
IMTL	0.21	0.15	0.40	0.31	−0.03
3_TDW	0.11	0.30	−0.25	0.19	−0.17
Eigenvalue	0.15	0.01	0.01	0.01	0.00
% variance	82.76	6.73	5.76	3.28	1.07

tarsus smooth, inner tarsal fold absent; tips of all toes slightly dilated, forming truncated disks, much wider than those of fingers (disk diameter of the third toe 0.8 mm; 3FDW/3TDW 0.7), disks of all toes with peripheral grooves, dorsal surface of toe disks II–V with short median longitudinal grooves; relative toe disk widths: I<IV<V<II<III; webbing between toes: preaxial side of toes II–IV, postaxial side of toe IV reaching distal subarticular tubercles, at all toes webbing reaching disk as fringe, webbing formula: I1–2II1–2½III1–2½IV2½–1V; subarticular tubercles on toes distinct, round, formula: 1 : 1 : 2 : 3 : 2 (for toes I : II : III : IV : V, respectively); two metatarsal tubercles: inner metatarsal tubercle elongated, prominent, outer metatarsal tubercle weak.

Dorsal skin slightly bumpy with low tubercles, scattered over dorsal surfaces of limbs; eyelid without supraciliary spines; supratympanic fold weakly developed, from posterior corner of eye to arm insertion; lateral side of head and flank smooth; ventral side of body and limbs smooth, vent area smooth.

Coloration of holotype in life. Dorsal surface of head and trunk greyish-brown to light-brown with dark-brown markings, a distinct dark-brown interorbital bar between eyelids, forming a reverse arrow in shape, running posteriorly towards scapular region and not covering dorsal surfaces of upper eyelids; one small black spot adjacent behind the eyes; a dorsal dark-brown marking consisting of two reverse V-shaped figures forming an hourglass-shaped marking: an anterior reverse V-shaped figure runs from scapular area posteriorly and laterally, opening at level of axilla and getting narrower again posteriorly on middle of dorsum; posterior reverse ↓-shaped figure starts at middle of dorsum and runs laterally towards groin; few small black scapular spots in the flank-belly and inguinal region, both usually bordered with a thin light-brown line; supratympanic fold light-brown; dorsolateral surfaces of trunk and upper arm brown with gray-pattern a few small black spots; dorsal surfaces of thigh, tibia, and tarsus brown with dark-brown cross-bars on each hindlimb; fingers and toes dorsally brown with dark-brown cross-bars; throat and chest white-grey with intense small dark-grey mottling, belly lighter with indistinct grayish mottling, limbs ventrally white-grey with small dark-grey mottling; hand and foot ventrally greyish-brown; pupil black, fine golden reticulations throughout iris.

Coloration of holotype in preservative. In ethanol, dorsal coloration changed to dark greyish-brown, ventral surface of chest, belly, and limbs greyish-beige; dorsal pattern, dark spots on dorsum and stripes on dorsal surfaces of limbs unchanged, dark brown pattern changed to dark grey; iris completely black.

Variation (Table 3, Figures 4A, 4B, 4C, 4D, 4E, 4F, 4G, 4H). Specimens vary in body size and dorsal pattern from dark

Figure 4 Paratypes of *Microhyla hongiaoensis* **sp. nov.** in life. A, male IEBR4575 and female IEBR4576 in amplexus; B, female VNMN 07390; C, female VNMN 07449; D, male VNMN 07449; E, male VNMN 07388; F, male IEBR4573; G, male IEBR4574; H, male CIB-VNMN 07617. Photos by Chung V. Hoang.

Figure 5 Habitat of *Microhyla hongiaoensis* **sp. nov.** in Bi Doup-Nui Ba National Park, Lam Dong Province, Vietnam. Photos by Chung V. Hoang.

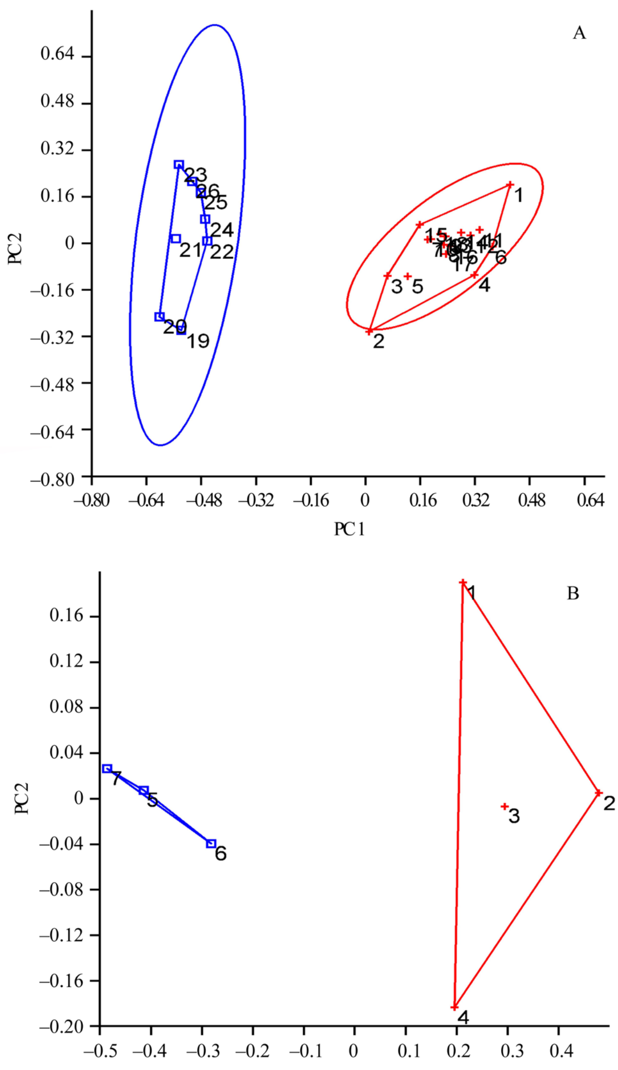

Figure 6 Plots of the first principal component (PC1) versus the second (PC2) *Microhyla hongiaoensis* **sp. nov.** (blue, blue 24A is the holotype) and topotype of *M. pulchella* (red, red 1A is the holotype). Males (A) and the females (B).

brown markings or with black scapular spots. All adult males have thumbs on anterior part of forearm but absent in adult females. Three female paratypes (VNMN 7390, 7447, CIB-VNMN 07448 have a larger body size than those of male paratypes (SVL 23.27 ± 1.64 mm [18.1–25.8 mm; n = 3] mm vs. 17.40 ± 1.86 mm [14.7–21.6 mm; n = 8] in males); the male VNMN 7385 has a dark brown background color with blackish-brown markings. Two male paratypes (IEBR 4573, 4574) have fewer dark spots in the axilla area compared to the holotype. The male (CIB-VNMN 07617) has an unsymmetrical hourglass-shaped marking. Three females (VNMN 7390, IEBR 4576, CIB-VNMN 07483) have a lighter greyish ventral surface compared to the holotype.

Natural history All specimens were collected at night, from 19:00 to 23:00 h, on the banks of a small temporary pond that was formed after heavy rain, along the sides of a recently constructed road (Figures 5A, 5B). Specimens of the new species were recorded at an elevation of ca. 1513 m a.s.l. Male and female frogs were found in amplexus on the water surface of the pond. Males were found calling about 1 m from the pond at night. The 27C road separated the Hon Giao forest into two patches. The new species was found sympatrically with five congeners, including *M. pineticola*, *M. mukhlesuri*, *M. annamensis*, *M. berdmorei*, *M. pulchella* (Figures 7A, 7B, 7C, 7D, 7E, 7F) and *Micryletta inornata*, all of which were reproducing simultaneously with the new species and sharing the same breeding site. Other anurans such as *Feihyla palpebralis* (Smith), *Polypedates* sp. (the *P. leucomystax* species complex), *Rhacophorus calcaneus* Smith, *Fejervarya limnocharis* (Gravenhorst), and *Occidozyga martensii* (Peters) also encountered in surroundings. Larval stages and eggs of the new species are unknown.

Distribution The new species currently known only from the type locality in Bi Doup-Nui Ba National Park, Lac Duong District, Lam Dong Province, Vietnam (Figure 1).

Conservation status The new species is likely to be endemic

Table 6 Selected diagnostic characters for the comparisons between the new species and *M. pulchella*.

Species	*M. hongiaoensis* **sp. nov.**	*M. pulchella*
Habitus	Slender	Moderately stocky
Dorsum skin	Scattered by small tubercles	Smooth
Dorsal median longitudinal grooves on finger disks	Absent	Present, weak
Dorsal peripheral grooves on toe disks	Present, weak T2-T5	Present, weak
Foot webbing	I1–2II1–2½III1–2½IV2½–1V	I1½–2II1–2III–2½IV2¼–1V
Two small black scapular spots on dorsum, both usually bordered by a thin white line	Absent	Present
Few black scapular spots behind the eyes	Absent	Present

Figure 7 The five sympatric species of *Microhyla* recorded at the type locality of *M. hongiaoensis* **sp. nov.** (Vietnam, Lam Dong Province, Bi Doup-Nui Ba NP). A, *M. pineticola* (SVL = 23.7 mm); B, *M. mukhlesuri* (SVL = 25.8 mm); C, *M. annamensis* (SVL= 18.7 mm); D, *M. berdmorei* (SVL = 33.4 mm) and E, *M. pulchella* (Female SVL = 23.9 mm); F, *M. pulchella* (Male SVL = 17.3 mm). Photos by Chung V. Hoang.

to Langbian Plateau, central Vietnam. However, the extent of its actual distribution range should be confirmed in further studies. Given the available information, we suggest this species be considered as Data Deficient following IUCN's Red List categories (IUCN 2001).

Etymology Specific epithet is in reference to the type locality of the Hon Giao forest. We recommend "Hongiao Narrow-mouth Frog" as the common English name and "Nhái bầu hòn giao" as the Vietnamese name.

4. Discussion

Our matrilineal genealogy was consistent with those of Matsui *et al.* (2011), Peloso *et al.* (2016), Nguyen *et al.* (2019) and Garg *et al.* (2019). The BI genealogy showed that monophyly of Microhylinae was not supported and the relationships among microhylid subfamilies remained unresolved (Nguyen *et al.*, 2019) (Figure 2). In our phylogenetic analyses, the newly collected *Microhyla* specimens were recovered as sister to *M. pulchella* (Poyarkov *et al.*, 2014) with high nodal support values (1.0/99.7). Furthermore, the results of morphometric analyses (PCA) indicated that *M. hongiaoensis* could be well separated from *M. pulchella* (Poyarkov *et al.*, 2014). Interestingly, both species were found sympatrically at the same habitat and elevation (ca. 1500 m.s.l) and they also share the same breeding site with other microhylid species in Hon Giao forest, Bi Doup-Nui Ba NP, Lam Dong Province, Langbian plateau, Vietnam. This gives us a hypothesis that during the formation of the two sister's species of *Microhyla*, the Liangbian plateau region had been strongly geographically divided for a long time. Later, the geographical barriers between the two species faded away and created common habitat nests for the two sister's species. However, further studies are required to prove this hypothesis.

The herpetofauna of Truong Son Range is well known in terms of species richness of local endemism with many new species have been discovered recently (eg. Hoang *et al.*, 2013; Inger *et al.*, 1999; Orlov *et al.*, 2005; Rowley *et al.*, 2010b, 2016; Ziegler *et al.*, 2008). The discovery of *M. hongiaoensis* brings the total number of known species in the genus *Microhyla* to 50 and the species number in Vietnam to 18 (Frost *et al.*, 2019). The Truong Son Range harbors the highest diversity of the genus *Microhyla* with 15 recorded species so far. However, habitat loss is one of the greatest threats to amphibians in Southeast Asia, and the amphibians of the region appear to be particularly vulnerable to habitat alterations (Rowley *et al.*, 2010a, 2016). The need for biological exploration in this region is made urgent due to intensifying logging, road construction, increasing agricultural pressure and other human activities (De Koninck 1999; Kuznetsov and Kuznetsova 2011; Laurance 2007; Meijer 1973; Meyfroidt and Lambin 2008).

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